

CENTER FOR DRUG EVALUATION AND RESEARCH

Approval Package for:

Application Number : 074544

Trade Name : LEUCOVORIN CALCIUM TABLETS USP

**Generic Name: Leucovorin Calcium Tablets USP 5mg (base)
and 25mg (base)**

Sponsor : Par Pharmaceutical, Inc.

Approval Date: August 28, 1997

CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION 074544

CONTENTS

	Included	Pending Completion	Not Prepared	Not Required
Approval Letter	X			
Tenative Approval Letter				
Approvable Letter				
Final Printed Labeling	X			
Medical Review(s)				
Chemistry Review(s)	X			
EA/FONSI				
Pharmacology Review(s)				
Statistical Review(s)				
Microbiology Review(s)				
Clinical Pharmacology				
Biopharmaceutics Review(s)				
Bioequivalence Review(s)	X			
Administrative Document(s)				
Correspondence				

CENTER FOR DRUG EVALUATION AND RESEARCH

Application Number **074544**

APPROVAL LETTER

AUG 28 1997

Par Pharmaceutical, Inc.
Attention: Michelle Bonomi
One Ram Ridge Road
Spring Valley, NY 10977

Dear Madam:

This is in reference to your abbreviated new drug application dated September 16, 1994, submitted pursuant to Section 505(j) of the Federal Food, Drug, and Cosmetic Act, for Leucovorin Calcium Tablets USP, 5 mg (base) and 25 mg (base).

Reference is also made to your amendment July 10, 1997.

We have completed the review of this abbreviated application and have concluded that the drug is safe and effective for use as recommended in the submitted labeling. Accordingly, the application is approved. The Division of Bioequivalence has determined your Leucovorin Calcium Tablets USP, 5 mg (base) and 25 mg (base) to be bioequivalent, and therefore, therapeutically equivalent to that of the listed drug, Wellcovorin® Tablets, 5 mg (base) and 25 mg (base), respectively, of Glaxo Wellcome, Inc.

Under 21 CFR 314.70, certain changes in the conditions described in this abbreviated application require an approved supplemental application before the change may be made.

Post-marketing reporting requirements for this abbreviated application are set forth in 21 CFR 314.80-81. The Office of Generic Drugs should be advised of any change in the marketing status of this drug.

We request that you submit, in duplicate, any proposed advertising or promotional copy which you intend to use in your initial advertising or promotional campaigns. Please submit all proposed materials in draft or mock-up form, not final print. Submit both copies together with a copy of the proposed or final printed labeling to the Division of Drug Marketing, Advertising, and Communications (HFD-240). Please do not use Form FD-2253 (Transmittal of Advertisements and Promotional Labeling for Drugs for Human Use) for this initial submission.

We call your attention to 21 CFR 314.81(b)(3) which requires that materials for any subsequent advertising or promotional campaign be submitted to our Division of Drug Marketing, Advertising, and Communications (HFD-240) with a completed Form FD-2253 at the time of their initial use.

Sincerely yours,

for
Douglas L. Sporn
Director
Office of Generic Drugs
Center for Drug Evaluation and Research

8/20/97

CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER **074544**

FINAL PRINTED LABELING



NDC 49884-238-15
**LEUCOVORIN
CALCIUM
TABLETS, USP**
25 mg*

CAUTION: Federal law prohibits
dispensing without prescription.
25 TABLETS

*Each tablet contains:
Leucovorin calcium equivalent to
25 mg leucovorin.

USUAL DOSAGE:
Read Accompanying Literature.
**KEEP THIS AND ALL DRUGS
OUT OF REACH OF CHILDREN.**
Dispense in a tight, light-resistant
container as defined in the USP.
Store between 15°-25°C (59°-77°F).
Protect from light and moisture.

Control No.:

Exp. Date:

Tablet #238
10295

Par Pharmaceutical, Inc.
Spring Valley, NY 10977



N 3 49884-238-15 5



NDC 49884-238-01
**LEUCOVORIN
CALCIUM
TABLETS, USP**
25 mg*

CAUTION: Federal law prohibits
dispensing without prescription.
100 TABLETS

*Each tablet contains:

Leucovorin calcium equivalent to
25 mg leucovorin.

USUAL DOSAGE:
Read Accompanying Literature.
**KEEP THIS AND ALL DRUGS
OUT OF REACH OF CHILDREN.**
Dispense in a tight, light-resistant
container as defined in the USP.
Store between 15°-25°C (59°-77°F).
Protect from light and moisture.

Control No.:

Exp. Date:

Tablet #238
10295

Par Pharmaceutical, Inc.
Spring Valley, NY 10977



N 3 49884-238-01 8



NDC 49884-237-01
**LEUCOVORIN
CALCIUM
TABLETS, USP**
5 mg*

CAUTION: Federal law prohibits
dispensing without prescription.
100 TABLETS

*Each tablet contains:

Leucovorin calcium equivalent to
5 mg leucovorin.

USUAL DOSAGE:
Read Accompanying Literature.
**KEEP THIS AND ALL DRUGS
OUT OF REACH OF CHILDREN.**
Dispense in a tight, light-resistant
container as defined in the USP.
Store between 15°-25°C (59°-77°F).
Protect from light and moisture.

Control No.:

Exp. Date:

Tablet #237
10295

Par Pharmaceutical, Inc.
Spring Valley, NY 10977



N 3 49884-237-01 1



NDC 49884-237-07
**LEUCOVORIN
CALCIUM
TABLETS, USP**
5 mg*

CAUTION: Federal law prohibits
dispensing without prescription.
20 TABLETS

*Each tablet contains:

Leucovorin calcium equivalent to
5 mg leucovorin.

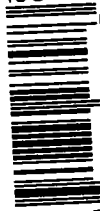
USUAL DOSAGE:
Read Accompanying Literature.
**KEEP THIS AND ALL DRUGS
OUT OF REACH OF CHILDREN.**
Dispense in a tight, light-resistant
container as defined in the USP.
Store between 15°-25°C (59°-77°F).
Protect from light and moisture.

Control No.:

Exp. Date:

Tablet #237
10295

Par Pharmaceutical, Inc.
Spring Valley, NY 10977



N 3 49884-237-07 3



NDC 49884-237-11
**LEUCOVORIN
CALCIUM
TABLETS, USP**
5 mg*

CAUTION: Federal law prohibits
dispensing without prescription.
30 TABLETS

*Each tablet contains:

Leucovorin calcium equivalent to
5 mg leucovorin.

USUAL DOSAGE:
Read Accompanying Literature.
**KEEP THIS AND ALL DRUGS
OUT OF REACH OF CHILDREN.**
Dispense in a tight, light-resistant
container as defined in the USP.
Store between 15°-25°C (59°-77°F).
Protect from light and moisture.

Control No.:

Exp. Date:

Tablet #237
10295

Par Pharmaceutical, Inc.
Spring Valley, NY 10977



N 3 49884-237-11 0



**LEUCOVORIN CALCIUM
TABLETS, USP**

105/97



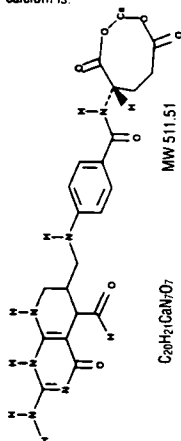
010237-01

SAMPLE

DESCRIPTION

Leucovorin calcium tablets contain 5 mg or 25 mg leucovorin as the calcium salt of *N*-[4-[[[(2-amino-5-formyl-1,4,5,6,7,8-hexahydro-4-oxo-6-pteridiny)-methyl]amino]benzoyl]-L-glutamic acid. This is equivalent to 5.4 mg or 27.01 mg of anhydrous leucovorin calcium. In addition, each tablet contains colloidal silicon dioxide, croscarmellose sodium, anhydrous lactose, magnesium stearate and microcrystalline cellulose.

Leucovorin is a water soluble form of reduced folate in the folate group; it is useful as an antidote to drugs which act as folic acid antagonists. These tablets are intended for oral administration only. The structural formula of leucovorin calcium is:



CLINICAL PHARMACOLOGY

Leucovorin is a racemic mixture of the diastereoisomers of the 5-formyl derivative of tetrahydrofolic acid. The biologically active compound of the mixture is the (-)-L-isomer, known as *Citrovorum factor*, or (-)-folinic acid. Leucovorin does *not* require reduction by the enzyme dihydrofolate reductase in order to participate in reactions utilizing folates as a source of "one-carbon" moieties. Following oral administration, leucovorin is rapidly absorbed and enters the general body pool of reduced folates. The increase in plasma and serum folate activity (determined microbiologically with *Lactobacillus casei*) seen after oral administration of leucovorin is predominantly due to 5-methyltetrahydrofolate.

quire reduction by the enzyme dihydrofolate reductase in order to participate in reactions utilizing folates as a source of "one-carbon" moieties. Following oral administration, leucovorin is rapidly absorbed and enters the general body pool of reduced folates. The increase in plasma and serum folate activity (determined microbiologically with *Lactobacillus casei*) seen after oral administration of leucovorin is predominantly due to 5-methyltetrahydrofolate.

Twenty normal men were given a single, oral 15 mg dose (7.5 mg/m²) of leucovorin calcium and serum folate concentrations were assayed with *L. casei*. Mean values observed (\pm one standard error) were:

- a) Time to peak serum folate concentration: 1.72 ± 0.08 hours.
- b) Peak serum folate concentration achieved: 268 ± 18 ng/mL.
- c) Serum folate half-disappearance time: 3.5 hours.

Oral tablets yielded areas under serum folate concentration-time curves (AUCs) that were 12% greater than equal amounts of leucovorin given intramuscularly and equal to the same amounts given intravenously.

Oral absorption of leucovorin is saturable at doses above 25 mg. The apparent bioavailability of leucovorin was 97% for 25 mg, 75% for 50 mg and 37% for 100 mg.

INDICATIONS AND USAGE

Leucovorin calcium tablets are indicated to diminish the toxicity and counteract the effects of impaired methotrexate elimination and of inadvertent overdosages of folic acid antagonists.

CONTRAINDICATIONS

Leucovorin is improper therapy for pernicious anemia and other megaloblastic anemias secondary to the lack of vitamin B₁₂. A hematologic remission may occur while neurological manifestations continue to progress.

WARNINGS

In the treatment of accidental overdosage of folic acid antagonists, leucovorin should be administered as promptly as possible. As the time interval between antifolate administration (e.g., methotrexate) and leucovorin rescue increases, leucovorin's effectiveness in counteracting hematologic toxicity decreases.

Monitoring of the serum methotrexate concentration is essential in determining the optimal dose and duration of treatment with leucovorin.

Delayed methotrexate excretion may be caused by a third space fluid accumulation (i.e., ascites, pleural effusion), renal insufficiency, or inadequate hydration. Under such circumstances, higher doses of leucovorin or prolonged administration may be indicated. Doses higher than those recommended for oral use must be given intravenously.

Leucovorin may enhance the toxicity of fluorouracil. Deaths from severe enterocolitis, diarrhea, and dehydration have been reported in elderly patients receiving weekly leucovorin and fluorouracil.¹ Concomitant granulocytopenia and fever were present in some but not all of the patients.

The concomitant use of leucovorin with trimethoprim-sulfamethoxazole for the acute treatment of *Pneumocystis carinii* pneumonia in patients with HIV infection was associated with increased rates of treat-

INDICATIONS AND USAGE

Leucovorin calcium tablets are indicated to diminish the toxicity and counteract the effects of impaired methotrexate elimination and of inadvertent overdoses of folic acid antagonists.

CONTRAINDICATIONS

Leucovorin is improper therapy for pernicious anemia and other megaloblastic anemias secondary to the lack of vitamin B₁₂. A hematologic remission may occur while neurological manifestations continue to progress.

WARNINGS

In the treatment of accidental overdosage of folic acid antagonists, leucovorin should be administered as promptly as possible. As the time interval between antifolate administration (e.g., methotrexate) and leucovorin rescue increases, leucovorin's effectiveness in counteracting hematologic toxicity decreases.

Monitoring of the serum methotrexate concentration is essential in determining the optimal dose and duration of treatment with leucovorin.

Delayed methotrexate excretion may be caused by a third space fluid accumulation (i.e., ascites, pleural effusion), renal insufficiency, or inadequate hydration. Under such circumstances, higher doses of leucovorin or prolonged administration may be indicated. Doses higher than those recommended for oral use must be given intravenously.

Leucovorin may enhance the toxicity of fluorouracil. Deaths from severe enterocolitis, diarrhea, and dehydration have been reported in elderly patients receiving weekly leucovorin and fluorouracil.¹ Concomitant granulocytopenia and fever were present in some but not all of the patients.

The concomitant use of leucovorin with trimethoprim-sulfamethoxazole for the acute treatment of *Pneumocystis carinii* pneumonia in patients with HIV infection was associated with increased rates of treatment failure and mortality in a placebo controlled study.

PRECAUTIONS

General: Parenteral administration is preferable to oral dosing if there is a possibility that the patient may vomit or not absorb the leucovorin. Leucovorin has no effect on other established toxicities of methotrexate, such as the nephrotoxicity resulting from drug and/or metabolite precipitation in the kidney.

Drug Interactions: Folic acid in large amounts may counteract the antiepileptic effect of phenobarbital, phenytoin and primidone, and increase the frequency of seizures in susceptible children.

Preliminary animal and human studies have shown that small quantities of systemically administered leucovorin enter the CSF primarily as 5-methyltetrahydrofolate and, in humans,

25 mg should be given parenterally (see **CLINICAL PHARMACOLOGY**).

Hydration (3L/d) and urinary alkalization with sodium bicarbonate should be employed concomitantly. The bicarbonate dose should be adjusted to maintain the urine pH at 7.0 or greater.

The recommended dose of leucovorin to counteract hematologic toxicity from folic acid antagonists with less affinity for mammalian dihydrofolate reductase than methotrexate (i.e., trimethoprim, pyrimethamine) is substantially less and 5 to 15 mg of leucovorin per day has been recommended by some investigators.

Patients who experience delayed early methotrexate elimination are likely to develop reversible non-oliguric renal failure. In addition to appropriate leucovorin therapy, these patients require continuing hydration and urinary alkalization, and close monitoring of fluid and electrolyte status, until serum methotrexate level has fallen to below 0.05 micromolar and the renal failure has resolved.

Some patients will have abnormalities in methotrexate elimination or renal function following methotrexate administration, which are significant but less severe. These abnormalities may or may not be associated with significant clinical toxicity. If significant clinical toxicity is observed, leucovorin rescue should be extended for an additional 24 hours (total of 14 doses over 84 hours) in subsequent courses of therapy. The possibility that the patient is taking other medications which interact with methotrexate (e.g., medications which may interfere with methotrexate elimination or binding to serum albumin) should always be reconsidered when laboratory abnormalities or clinical toxicities are observed.

HOW SUPPLIED

Leucovorin calcium tablets 5 mg are off white, round tablets, containing 5 mg leucovorin as the calcium salt, debossed "Par 237" with bisect on one side and plain on the other and are available in bottles of 20 (NDC 49884-237-07), 30 (NDC 49884-237-11) and 100 (NDC 49884-237-01).

Leucovorin calcium tablets 25 mg are off white, round tablets, containing 25 mg leucovorin as the calcium salt, debossed "Par 238" with bisect on one side and plain on the other and are available in bottles of 25 (NDC 49884-238-15) and 100 (NDC 49884-238-01).

Store between 15° - 25°C (59° - 77°F). Protect from light and moisture.

CAUTION: Federal law prohibits dispensing without prescription.

REFERENCES

1. Grem JL, Shoemaker DD, Petrelli NJ, Douglass HO Jr. Severe and fatal toxic effects observed in treatment with high- and low-dose leucovorin plus 5-Fluorouracil for colorectal carcinoma. *Cancer Treat Rep* 1987; 71: 1122.
2. Link MP, Goorin AM, Miser AW et al. The effect of adjuvant chemotherapy on relapse-free survival patients with osteosarcoma of the extremity. *N Engl J Med* 1986; 314: 1600-1606.

Manufactured by:
PAR PHARMACEUTICAL, INC.
Spring Valley, NY 10977
Issued: 05/97

4

remain 1 to 3 orders of magnitude lower than the usual methotrexate concentrations following intrathecal administration. However, high doses of leucovorin may reduce the efficacy of intrathecally administered methotrexate.

Leucovorin may enhance the toxicity of fluorouracil (see **WARNINGS**).

Pregnancy: Teratogenic Effects: Pregnancy Category C. Animal reproduction studies have not been conducted with leucovorin. It is also not known whether leucovorin can cause fetal harm when administered to a pregnant woman or can affect reproduction capacity. Leucovorin should be given to a pregnant woman only if clearly needed.

Nursing Mothers: It is not known whether this drug is excreted in human milk. Because many drugs are excreted in human milk, caution should be exercised when leucovorin is administered to a nursing mother.

Pediatric Use: See Drug Interactions subsection.

ADVERSE REACTIONS

Allergic sensitization, including anaphylactoid reactions and urticaria, has been reported following the administration of both oral and parenteral leucovorin.

OVERDOSAGE

Excessive amounts of leucovorin may nullify the chemotherapeutic effect of folic acid antagonists.

DOSAGE AND ADMINISTRATION

Leucovorin calcium tablets are intended for oral administration. Because absorption is saturable, oral administration of doses greater than 25 mg is not recommended.

Impaired Methotrexate Elimination or Inadvertent Overdosage: Leucovorin rescue should begin as soon as possible after an inadvertent overdosage and within 24 hours of methotrexate administration when there is delayed excretion (see **WARNINGS**)². Leucovorin 15 mg (10 mg/m²) should be administered IM, IV, or PO every 6 hours until serum methotrexate level is less than 10⁻⁶M. In the presence of gastrointestinal toxicity, nausea or vomiting, leucovorin should be administered parenterally.

Serum creatinine and methotrexate levels should be determined at 24 hour intervals. If the 24 hour serum creatinine has increased 50% over baseline or if the 24 hour methotrexate level is greater than 5 x 10⁻⁶M or the 48 hour level is greater than 9 x 10⁻⁷M, the dose of leucovorin should be increased to 150 mg (100 mg/m²) IV every 3 hours until the methotrexate level is less than 10⁻⁶M. Doses greater than 25 mg should be given parenterally (see **CLINICAL PHARMACOLOGY**).

Hydration (3L/d) and urinary alkalinization with sodium bicarbonate should be employed concomitantly. The bicarbonate dose should be adjusted to maintain the urine pH at 7.0 or greater.

The recommended dose of leucovorin to counteract hematologic toxicity from folic acid antagonists with less affinity for mammalian dihydrofolate reductase than methotrexate (i.e.,

CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER 074544

CHEMISTRY REVIEW(S)

1. CHEMISTRY REVIEW NO.5
2. ANDA # 74-544
3. NAME AND ADDRESS OF APPLICANT
Par Pharmaceutical, Inc.
One Ridge Road
Spring Valley, NY 10977
4. BASIS OF SUBMISSION
Acceptable per CR # 1.
5. SUPPLEMENT(s)
N/A
6. PROPRIETARY NAME
None Used
7. NONPROPRIETARY NAME
Leucovorin Calcium Tablets USP
8. SUPPLEMENT(s) PROVIDE(s) FOR:
N/A
9. AMENDMENTS AND OTHER DATES:
FIRM:
Original Submission: 9-16-94
Major Amendment: 4-10-95 (Response to NA letter dated 2-17-95
Amendment 5-31-95 (Bio)
Minor Amendment: 8-17-95 (Response to NA letter dated 7-24-95).
NC:12-29-95 (Response to bio's letter dated 12-7-95)
Minor Amendment: 1-30-96 (Response to 1-23-96 NA letter)
Bio Data: 2-15-96
Bio Data: 3-12-96
* Minor Amendment: 7-10-97

FDA:
Accepted for filing on 9-20-94
Acknowledgment Letter: 10-6-94
NA Letter (Chemistry + Labeling): 2-17-95
BIO deficiency letter: 4-28-95
NA letter (Chemistry + Labeling): 7-24-95
NA letter (BIO): 12-7-95
NA letter : 1-23-96 (based on bio deficiencies)
Bio Acceptance letter: 4-19-96
NA letter (CGMP issue): 6-13-96
Labeling revision request letter: 4-30-97
10. PHARMACOLOGICAL CATEGORY
For treatment of undesired hematopoietic effects of folic acid antagonists.

11. Rx or OTC
Rx

12. RELATED IND/NDA/DMF(s)

13. DOSAGE FORM
Tablets

14. POTENCY
5 mg & 25 mg

15. CHEMICAL NAME AND STRUCTURE

NAME:

Calcium Salt of N-[4-[[2.amino-5-formyl-1,4,5,6,7,8-hexahydro-4-oxo-6-pteridiny]methyl]amino]-L-glutamic acid

STRUCTURE: Listed in labeling insert per USP 23.

16. RECORDS AND REPORTS
N/A

17. COMMENTS

Par has fulfilled all the requirements for an approval from chemistry and labeling point of view again. Approval letter went through administration review after completion of CR # 4 dated 4-16-96. But A NA letter was sent to the firm on 6-13-96 based on cGMP issue for manufacturer.

18. CONCLUSIONS AND RECOMMENDATIONS
Approved pending acceptable EER.

19. REVIEWER: DATE COMPLETED:
Mujahid L. Shaikh 7-29-97

ANDA 74-544
Dup File
Division File
Field Copy

Endorsements:

HFD-625/MShaikh/7-29-97
HFD-625/MSmela/7-30-97
x:\new\firmnsz\par\ltrs&rev\74544REV.5
F/t by: bc/7-30-97

- 7/31/97

- 8/1/97

CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER 074544

BIOEQUIVALENCE REVIEW(S)

ANDA 74-544

DEC - 1995

Par Pharmaceutical, Inc.
Attention: Diana Sloane
One Ram Ridge Road
Spring Valley NY 10977

Dear Madam:

Reference is made to the Abbreviated New Drug Application submitted on May 31, 1995, for Leucovorin Calcium Tablets USP, 5 mg (Eg. base) and 25 mg (Eg. base).

The Office of Generic Drugs has reviewed the bioequivalence data submitted and the following comments are provided for your consideration:

1. The data presented on page 1023 (diskette file) and on page 965 in the study summary data tables do not match with regard to AUC_{INF} values (with three exceptions) and for eight of the AUC_{0-t} entries. Please explain these discrepancies and submit another 3.5" diskette with the correct AUC_{0-t} values and revised AUC_{INF} values. It is not necessary to include log-transformed values in the diskette file.
- 2.

As described under 21 CFR 314.96 an action which will amend this application is required. The amendment will be required to address all of the comments presented in this letter. Should you have any questions, please call Jason A. Gross, Pharm.D., at (301) 594-2290. In future correspondence regarding this issue, please include a copy of this letter.

Sincerely yours,

✓ Keith K. Chan, ~~Ph.D.~~
Director, Division of Bioequivalence
Office of Generic Drugs
Center for Drug Evaluation
and Research

OCT 24 1995

Leucovorin Calcium
25 & 5 mg tablets
ANDA #74-544
Reviewer: James D. Henderson
File: 74544SDW.994 595

Par Pharmaceuticals
Spring Valley, NY
Submitted:
May 31, 1995

RESPONSE TO REVIEW OF A BIOEQUIVALENCE STUDY

I. Background

On 9/16/94 the sponsor submitted the results of a bioequivalence study comparing its test product leucovorin calcium 25 mg tablets with the reference listed drug (RLD) Wellcovorin® (Burroughs Wellcome, NDA #18-342, 7/8/83; received 1/4/95). In addition, the sponsor requested waiver of in vivo bioequivalence study requirements for the 5 mg strength of the test product based on formula proportionality and dissolution data. The study was reviewed and found incomplete (file date 3/31/95). In the present submission, the sponsor has responded to the deficiency comments in the letter of 4/28/95.

II. Responses to Deficiency Comments

1. On p. 95 and 429 the test product lot# is stated as "SB026". On p. 169, 193-4, 438, and 440-1, the test product lot# is stated as "SB0026". The sponsor should state the correct lot number for the test biostudy lot.

Sponsor's Response: Correct batch # is SB0026.

Reviewer's Comment: Acceptable.

2. The Division of Bioequivalence guidance for leucovorin calcium tablets requests that the sponsor determine the ratio of _____ in both the test and reference biostudy lots, and that these ratios should not differ by more than _____. This information was not submitted.

Reviewer's Comment: Acceptable

3. For the dissolution data, the sponsor did not provide CV's for the mean dissolution values at each sampling time or the analytical method.

Sponsor's Response: Requested data is provided.

Reviewer's Comment: The revised dissolution data is shown in Table 1.

4. The sponsor must provide case report forms, adverse reaction reports, and all other relevant clinical raw data from the study.

Sponsor's Response: Requested data is provided.

Reviewer's Comment: The reviewer inspected the submitted Case Report Forms with regard to inclusion/exclusion criteria, concomitant medications, and adverse events.

- Adverse Events: From the Raw Data, the reviewer found eight events reported. Two events were unrelated to the drug or study procedures, and the other six events (headache, 2; tired, 2; irritable, 1; spacey, 1) were judged as unlikely to be drug-related. All events were judged not serious and of mild intensity, and required no action.
- Inclusion/Exclusion Criteria: In two cases, subjects (S21, S32) were included despite a childhood history of asthma (last attack age 12). In two cases, subjects (S30, S32) were included despite history of hepatitis (full recovery in 1990).
- There were no concomitant medications taken during the study.

5.

Sponsor's Response: KEL values were recalculated and a revised PK report is submitted.

Reviewer's Comment: The sponsor submitted a diskette that should have contained the revised pharmacokinetic data (recommendation #2 from the previous review). Using the data from the diskette

with no modifications and the GLM procedure of SAS, the reviewer attempted to confirm the 90% CI results reported by the sponsor. For AUCINF the sponsor reported a 90% CI of 89.6-105.6 (p. 983); however, using the diskette data the reviewer obtained 89.7-105.8. Inspection of the data from the diskette (p. 1023) and the data tables (p. 965-6) shows that virtually all of the AUCINF values are different (with three exceptions) and that 8 of the AUC0-t values are different.

Sponsor's Response:

¹ Shah VP, et al. Analytical methods validation: bioavailability, bioequivalence, and pharmacokinetic studies. J Pharm Sci 1992;81:309-12.

III. Deficiencies

1. The data presented on p. 1023 (diskette file) and on p. 965 in the study summary data tables do not match with regard to AUCINF values (with three exceptions) and for eight of the AUC0-t entries. Please explain these discrepancies and submit another 3.5" diskette with the correct AUC0-t values and revised AUCINF values. It is not necessary to include log-transformed values in the diskette file.
2. The response to deficiency comment #8 is unacceptable. If

IV. Recommendations

1. The bioequivalence study conducted by Par Pharmaceutical on its leucovorin calcium 25 mg tablet, lot #SB0026, comparing it to Burroughs Wellcome's Wellcovorin® 25 mg tablet, lot #2T2271, has been found incomplete by the Division of Bioequivalence due to deficiencies 1-2 stated above.
2. The firm should be informed of deficiencies #1-2 and recommendation #1.

✓
James D. Henderson, Ph.D.
Review Branch II
Division of Bioequivalence

RD INITIALED RPAITNAIK
FT INITIALED RPAITNAIK

A

10/24/95

Concur. _____ Date _____
Keith K. Chan, Ph.D.
Director
Division of Bioequivalence

Table 1. In Vitro Dissolution Testing

Drug (Generic Name): leucovorin calcium
Dose Strength: 5 & 25 mg
ANDA No.: 74-544
Firm: Par
Submission Date: 5/31/95
File Name: 74544SDW.595

I. Dissolution Testing (USP Method):

USP XXII Basket: Paddle: X RPM: 50
No. Units Tested: 12
Medium: water Volume: 900 mL
Specifications: NLT 30 min
Reference Drug: Wellcovorin® (Burroughs Wellcome)
Assay Methodology:

II. Results of In Vitro Dissolution Testing:

Sampling Times (Minutes)	Test Product Lot #SB0027 Strength (mg) 5			Reference Product Lot #1W1638 exp 11/94 Strength (mg) 5		
	Mean %	Range	%CV	Mean %	Range	%CV
15	88.5		7.4	94.5		3.3
30	96.2		1.2	99.3		0.8
45	97.0		1.4	97.0		0.8
60	97.6		1.5	97.3		0.9

Sampling Times (Minutes)	Test Product Lot #SB0026 Strength (mg) 25			Reference Product Lot #2T2271 Strength (mg) 25 exp 8/95		
	Mean %	Range	%CV	Mean %	Range	%CV
15	95.9		2.2	97.5		2.6
30	98.7		1.3	99.3		1.2
45	99.5		1.0	99.6		0.9
60	100.0		1.0	99.1		0.7

ANDA 74-544

Par Pharmaceutical, Inc.
Attention: Michelle Bonomi
One Ram Ridge Road
Spring Valley NY 10977
|||||

APR - 9 1996

Dear Madam:

Reference is made to your abbreviated new drug application submitted pursuant to Section 505 (j) of the Federal Food, Drug and Cosmetic Act for Leucovorin Calcium Tablets USP, 5 mg (base) and 25 mg (base).

The Division of Bioequivalence has completed its review and has no further questions at this time.

Please note that the bioequivalency comments expressed in this letter are preliminary. The above bioequivalency comments may be revised after review of the entire application, upon consideration of the chemistry, manufacturing and controls, microbiology, labeling or other scientific or regulatory issues. A revised determination may require additional information and/or studies, or may conclude that the proposed formulation is not approvable.

Sincerely yours,

Keith K. Chan, Ph.D.
Director, Division of Bioequivalence
Office of Generic Drugs
Center for Drug Evaluation and Research

OFFICE OF GENERIC DRUGS
DIVISION OF BIOEQUIVALENCE

ANDA/AADA #74-544

SPONSOR: PAR

DRUG: LEUCOVORIN CALCIUM

DOSAGE FORM: TABLET

STRENGTHS/(s): 25 mg, 5 mg

TYPE OF STUDY: SINGLE DOSE, FASTING

STUDY SITE

STUDY SUMMARY:

1. Thirty-two subjects were enrolled in order to ensure completion of 30 subjects. The protocol stated that serum samples from all completed volunteers would be assayed. All 32 subjects completed the study.

2. The original submission and four subsequent amendments contained inconsistencies in the reported data. The sponsor explained the reasons for these inconsistencies and corrected the data.

3. Five analytical runs (LEU_004, LEU_007, LEU_113, LEU_020, LEU_023) were unacceptable based on QC sample results. This resulted in exclusion of all analytical data for Subjects 7, 8, 13, 14, 25, 26, 31, and 32. In addition, S15, Per. 2, and S30, Per. 1, predose samples were lost and the data could not be baseline-adjusted.

4. This leaves 22 of the 32 subjects for BE determination. The 90% CI's reported by the sponsor and confirmed by the reviewer were: logAUC_{0-t}, 90.8-111.0; logAUC_{INF}, 91.1-110.0; logC_{MAX}, 89.0-112.8.

5. The RATIO (AUC_{0-t}/AUC_{INF}) values obtained by the reviewer indicate that $\geq 85\%$, on average, of the total AUC is being captured during the sampling period. The results for RATIO and the 24-hr concentration values also indicate sufficient assay sensitivity. At 24 hr, the mean concentrations for test and reference treatments were 11.2 and 10.9 times the LOQ, respectively. However, the results for DURATION clearly indicate that sampling was stopped too soon since only 4 of 44 values were $> 3 \times 5\text{-MTHF}$ half-lives.

21 CFR 320.26(c)(ii) states that, in a single dose bioequivalence study, "...blood samples should be taken with a sufficient frequency to permit an estimate..." (ii) The total area under the curve for a time period at least three times the half-life of the active drug ingredient...". The failure to meet this criterion appears to be caused by the sponsor's strict adherence to the DBE Guidance which specified blood sampling up to 24 hours.

6. Since the RATIO mean values were > 0.8 for each treatment and assay

sensitivity appears adequate, the reviewer would recommend approval of the study.

WAIVER/DISSOLUTION:

The request for waiver of in vivo biostudy requirements for the 5 mg strength of the test product should be granted on the basis of acceptability of the 25 mg strength biostudy, acceptable in vitro dissolution testing results (file date 10/24/95), and similar proportionally formulas (file date 3/31/95). The formulation for the 5 mg strength is proportionally similar with respect to active ingredient and identical (except for filler) with respect to inactive ingredients (as a % of core weight) to the 25 mg strength of the test product that underwent bioequivalency testing.

PRIMARY REVIEWER: James D. Henderson, Ph.D. **BRANCH:** II
INITIAL: _____ **DATE** 3-26-96

BRANCH CHIEF: Rabindra N. Patnaik, Ph.D. **BRANCH:** II
INITIAL: _____ **DATE** 3/31/96

DIRECTOR, DIVISION OF BIOEQUIVALENCE:

Keith K. Cha-

INITIAL: _____ **DATE** 4/4/96

APR 1 1996

Leucovorin Calcium
25 & 5 mg tablets
ANDA #74-544
Reviewer: James D. Henderson
File: 74544SDW.D95

Par Pharmaceuticals
Spring Valley, NY
Submitted:
December 29, 1995 &
February 15, 1996 &
March 12, 1996

SUMMARY

1. Bioequivalence Review No.: 3

Review No. 1: Original submission 9/16/94, found incomplete file date 3/31/95

Review No. 2: Amendment submitted 5/31/95, found incomplete file date 10/24/95

2. Dates:

APPLICANT	FDA
Original Submission 9/16/94	Received by Reviewer 1/4/95
	RD Submitted 1/25/95
	RD Approved 3/31/95
	Final Submitted 3/31/95
	Final Approved 3/31/95
	Letter (Final) 4/28/95
Amendment 5/31/95	Received by Reviewer 9/8/95
	RD Submitted 9/21/95
	RD Approved 9/25/95
	Final Submitted 10/17/95
	Final Approved 10/24/95
	Letter (Final) 12/7/95
Amendment 12/29/95	Received by Reviewer 2/1/96
	Telecon Request 2/6/96
Amendment 2/15/96	Received 2/22/96
	Telecon Request 2/22/96
Amendment 3/12/96	Received 3/21/96
	RD Submitted 3/25/96

	RD Approved 3/26/96
	Final Submitted 3/26/96

3. Conclusions: Acceptable

4. Recommendations:

a. The bioequivalence study conducted by Par Pharmaceuticals on its leucovorin calcium 25 mg tablet, lot #SB0026, comparing it to Wellcovorin® 25 mg tablet has been found acceptable by the Division of Bioequivalence. The study demonstrates that Par's leucovorin calcium 25 mg tablet is bioequivalent to the reference product Wellcovorin® 25 mg tablet manufactured by Burroughs Wellcome.

b. The dissolution testing conducted by Par on its leucovorin calcium 25 mg tablet, lot #SB0026, is acceptable and should be incorporated into the firm's manufacturing controls and stability program. The dissolution testing should be conducted in 900 mL of water at 37° using USP 23 apparatus II (paddle) at 50 rpm. The test product should meet the following specifications:

Not less than of the labeled amount of the drug in the dosage form is dissolved in 30 minutes.

c. The dissolution testing conducted by Par on its leucovorin calcium 5 mg tablet, lot #SB0027, is acceptable. The firm has conducted an acceptable in vivo bioequivalence study (submitted 9/16/94 and 5/31/95) comparing the 25 mg tablet of the test product with the 25 mg tablet of the reference product Wellcovorin® manufactured by BW. The formulation for the 5 mg strength is proportionally similar with respect to active ingredient and identical (except for filler) with respect to inactive ingredients (as a % of core weight) to the 25 mg strength of the test product that underwent bioequivalency testing. The waiver of in vivo bioequivalence study requirements for the 5 mg strength of the test product is granted. The 5 mg tablet of the test product is therefore deemed bioequivalent to the 5 mg tablet of Wellcovorin® manufactured by BW.

d. From the bioequivalence point of view, the firm has met the requirements of in vivo bioequivalence and in vitro dissolution testing and the application is acceptable.

5. Signature Blocks and Routing:

U
James D. Henderson, Ph.D.
Review Branch II
Division of Bioequivalence

RD INITIALED RPAATNAIK
FT INITIALED RPAATNAIK

3/31/96

Concur: _____ Date 4/4/96
Keith K. Chan, Ph.D.
Director
Division of Bioequivalence

JDH/gj/3-26-96/74544

cc: ANDA #74-544 (original, duplicate), HFD-600 (Hare), HFD-630,
HFD-344 (CViswanathan), HFD-655 (Patnaik, Henderson), Drug
File, Division File

RESPONSES TO DEFICIENCY COMMENTS

Deficiency Comment #1:

The data presented on p. 1023 (diskette file) and on p. 965 in the study summary data tables do not match with regard to AUCINF values (with three exceptions) and for eight of the AUC0-t entries. Please explain these discrepancies and submit another 3.5" diskette with the correct AUC0-t values and revised AUCINF values. It is not necessary to include log-transformed values in the diskette file.

Sponsor's Response: The correct AUC0-t and AUCINF values were

•

•

Deficiency Comment #2:

ADDITIONAL COMMENTS

STUDY SUMMARY AND CONCLUSIONS

1. Thirty-two subjects were enrolled in order to ensure completion of 30 subjects. The protocol stated that serum samples from all completed volunteers would be assayed. All 32 subjects completed the study.
 2. The original submission and four subsequent amendments have contained inconsistencies in the reported data. The sponsor has now explained the reasons for these inconsistencies and corrected the data.
 3. Five analytical runs (LEU_004, LEU_007, LEU_113, LEU_020, LEU_023) were unacceptable based on QC sample results. This resulted in exclusion of all analytical data for Subjects 7, 8, 13, 14, 25, 26, 31, and 32. In addition, S15, Per. 2, and S30, Per. 1, predose samples were lost and the data could not be baseline-adjusted.
 4. This leaves 22 of the 32 subjects for BE determination. The 90% CI's reported by the sponsor and confirmed by the reviewer were: logAUC0-t, 90.8-111.0; logAUCINF, 91.1-110.0; logCMAX, 89.0-112.8.
 5. The RATIO values obtained by the reviewer indicate that \geq 85%, on average, of the total AUC is being captured during the sampling period. The results for RATIO and the 24-hr concentration values also indicate sufficient assay sensitivity. At 24 hr, the mean concentrations for test and reference treatments were 11.2 and 10.9 times the LOQ, respectively. However, the results for DURATION clearly indicate that sampling was stopped too soon since only 4 of 44 values were > 3 5-MTHF half-lives.
- 21 CFR 320.26(c)(ii) states that, in a single dose bioequivalence study, "...blood samples should be taken with a sufficient

frequency to permit an estimate...(ii) The total area under the curve for a time period at least three times the half-life of the active drug ingredient...". The failure to meet this criterion appears to be caused by the sponsor's strict adherence to the DBE Guidance which specified blood sampling up to 24 hours.

6. Since the RATIO mean values were > 0.8 for each treatment and assay sensitivity appears adequate, the reviewer would recommend approval of the study.

7. The request for waiver of in vivo biostudy requirements for the 5 mg strength of the test product should be granted on the basis of acceptability of the 25 mg strength biostudy, acceptable in vitro dissolution testing results (file date 10/24/95), and similar proportionally formulas (file date 3/31/95).

prepared by

3-26-96

James D. Henderson, Ph.D.
Review Branch II
Division of Bioequivalence

Table 1 - Additional Pharmacokinetic Parameters

<u>N</u>	<u>Parameter</u>	<u>Test</u>	<u>Reference</u>
22	RATIO ¹		
	Mean	0.8613	0.8580
	CV%	5.7	7.35
	Range	0.753-0.962	0.725-0.946
	DURATION ²		
	Mean	2.037	2.024
	CV%	30.7	33.2
	Range	1.35-4.12	0.988-3.23
	WASHOUT ³		
	Mean	28.51	28.34
	CV%	30.7	33.2
	Range	18.9-57.8	13.8-45.3
14 ⁴	RATIO ¹		
	Mean	0.8779	0.8803
	CV%	5.3	6.9
	Range	0.815-0.962	0.725-0.946
	DURATION ²		
	Mean	2.215	2.254
	CV%	30.1	32.5
	Range	1.57-4.12	0.988-3.23
	WASHOUT ³		
	Mean	31.01	31.5
	CV%	30.1	32.5
	Range	22.0-57.8	13.8-45.3

¹ RATIO = AUC_{0-t}/AUC_{INF}

² DURATION = $TLAST/t_{1/2}$

³ WASHOUT = $336/t_{1/2}$

⁴ Revised λ_z estimates with $R^2 > 0.9$

MAR 31 1995

Leucovorin Calcium
25 & 5 mg tablets
ANDA #74-544
Reviewer: James D. Henderson
File: 74544SDW.994

Par Pharmaceuticals
Spring Valley, NY
Submitted:
September 16, 1994

REVIEW OF A BIOEQUIVALENCE STUDY, DISSOLUTION DATA, AND WAIVER REQUEST

I. Background

The sponsor has submitted the results of a bioequivalence study comparing its test product leucovorin calcium 25 mg tablets with the reference listed drug (RLD) Wellcovorin® (Burroughs Wellcome, NDA #18-342, 7/8/83). In addition, the sponsor is requesting waiver of in vivo bioequivalence study requirements for the 5 mg strength of the test product based on formula proportionality and dissolution data. The application was received by the reviewer on 1/4/95.

On 8/4/88 the DBE issued a revised guidance for conducting bioequivalence studies of leucovorin calcium tablets. In order to seek approval of 25 and 5 mg tablets, an acceptable biostudy must be conducted comparing the 25 mg test product with Wellcovorin® 25 mg tablets, and a waiver from biostudy requirements may be requested for the 5 mg strength based on acceptable dissolution data and formula proportionality to the 25 mg strength. Assay of the metabolite 5-methyl-tetrahydrofolic acid may be by microbiological (measuring 5-formyl-THF plus 5-methyl-THF), HPLC, or RIA methods, but in all cases, endogenous folates must be measured at time zero, and later serum concentrations corrected. Both the potency and the ratio of D:L isomers of the test product biostudy lot should be within $\pm 5\%$ of the reference lot.

II. Study Site

Clinical and Analytical Site:

Principal Investigator:

Protocol #: 237-11, 5/3/94, IRB approval 5/3/94; amended 5/6/94, IRB approval 5/6/94

Study #: 16398

Study Dates: Period 1, 5/20-21/94 (dosing on 5/21); Period 2, 6/3-4/94 (dosing on 6/4/94)

Analytical Director:

Analysis Dates: 6/10-7/5/94

III. Study Design

This study was a randomized, single dose, two-treatment crossover design in 30 healthy male volunteers under fasting conditions

comparing equal doses the test product leucovorin calcium 25 mg tablets (1 X 25 mg) with the reference product Wellcovorin® (1 X 25 mg) with a 14-day washout period. Serum samples were assayed for the metabolite 5-methyltetrahydrofolic acid (5-MTHF) using a

IV. Subject Selection

Thirty-two healthy male volunteers were enrolled after giving informed consent to ensure completion of at least 30 subjects according to the following criteria:

A. Inclusion

- 19-50 years old
- good health as evidenced by medical history, physical examination, and laboratory tests (hematology, serum chemistry, urinalysis, HIV antibody screen, urine screen)
- body weight within $\pm 10\%$ of ideal weight for height and frame (Metropolitan Life Insurance Company, 1983)
- no clinically significant findings on physical examination
- normal laboratory values, unless the PI deems the abnormality as not clinically significant
- negative urine screen for alcohol and drugs of abuse

B. Exclusion

- history of alcohol or drug addiction within the last two years
- history of organ, systemic, or mental disease
- history of folate drug therapy during the past two years
- history of allergy or adverse reaction to leucovorin, folic acid, or related drugs
- participation within a clinical trial, blood donation of one pint or more, or treatment with any known enzyme-altering agent during the past 30 days
- plasmapheresis within 7 days prior to the study
- abnormal nutritional status
- use of any medication on a regular basis

V. Study Procedures

Treatments:

After an overnight fast of at least 10 hours, subjects received one of the following treatments:

1) Treatment A, leucovorin calcium (Par), 1 X 25 mg tablet, lot #SB0026, assay 95.9%; theoretical batch size finished size manufacturing date 11/5/93

2) Treatment B, Wellcovorin® (Burroughs Wellcome), 1 X 25 mg tablet, lot #2T2271, exp 8/95, assay 95.7%

Each dose was administered with 240 mL of water. After a 14-day washout, each subject was crossed over to the alternative treatment.

Restrictions:

- no Rx medication for at least 14 days and no OTC medication for 72 hours prior to and during the study
- no alcoholic beverages, caffeine/xanthine-containing foods, or foods high in folic acid for 48 hours prior to or during each study period
- confinement from the evening at least 10 hours prior to dosing until after the 24-hour sample
- no strenuous exercise during the confinement period
- may not lie down for the first 4 hours postdose

Meals and Fluids:

- fasting from 10 hours prior to dosing and for 4 hours postdose
- folate-rich foods will be excluded from the standardized meals
- water allowed freely except for 1 hour prior to dosing and 2 hours postdose

Blood Sampling:

Blood samples (10 mL) were collected into serum vacutainer tubes at 0 (predose), 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 8, 10, 12, 16, and 24 hours postdose, and allowed to clot at room temperature. Serum was separated by centrifugation (2500 rpm for 10 minutes at 4°). Sodium ascorbate solution (0.1 mL of a 10% solution per mL of serum) was added as antioxidant (final concentration 10 mg ascorbate/mL). Samples were stored within 90 minutes of collection at -70° until assayed.

VI. Analytical Methods and Data Analysis

A. Analytical Method (not for release under FOI)

B. Data Analysis

Pharmacokinetic variables were calculated from the concentration-time data: area under the curve to the time of the last measurable concentration (AUC_{0-t}) from trapezoidal integration; area under the curve extrapolated to infinity ($AUC = AUC_{0-t} + C_t/KEL$); peak drug concentration (C_{MAX}); time of peak drug concentration (T_{MAX}); terminal elimination rate constant (KEL) and half-life ($t_{1/2}$). These variables and their log-transformed values were analyzed by ANOVA (SAS v. 6.08 GLM procedure) using a general linear model containing factors for sequence, subjects within sequence, period, and formulation. 90% confidence intervals (CI) for the ratio of test and reference means were constructed for AUC's and C_{MAX}.

Pharmacokinetic analysis was performed on baseline-adjusted 5MTHF concentrations. The predose 5MTHF concentration was used as the baseline value and subtracted from all subsequent sample concentrations. An adjusted concentration less than zero was set to zero for analysis. Samples with concentrations below the LLOQ were set to zero for the AUC calculation; missing or nonreportable samples were not included in the trapezoidal area calculations. KEL was determined by linear regression of the $\ln(\text{concentration})$ vs. time data pairs for the linear terminal portion of the profile, and $t_{1/2} = 0.693/KEL$.

VII. Results

A. Product Information

1. Formulation: Table 1.
2. Potency: The potencies of the biostudy lots of test and reference products are within 5%.
3. Dissolution: Table 2.
4. Ratio of D:L isomers: not reported

B. Clinical

1. Completion: All 32 of the enrolled subjects completed the study. Samples from all of the 32 subjects were assayed and the results reported.

2. Adverse Events: There were seven adverse events in seven subjects. The severity of all of these events was judged as mild and unlikely or unrelated to the study medication: irritable (1), headache (2), tired (2), right shoulder pain (1), broken finger (1).

C. Pharmacokinetics/Statistics

1. Mean reported baseline-adjusted serum concentrations of 5MTHF are shown in Table 3. All subjects had serum concentrations > LLOQ over the entire 24-hour sampling interval for both treatments; therefore, $AUC_{0-t_{LAST}}$ is identical to AUC_{0-24} . There were no cases where the first nonzero concentration was the C_{MAX} .

2. Mean reported baseline-adjusted pharmacokinetic parameters of 5MTHF are shown in Table 4. The sponsor's analysis shows the absence of any statistically significant sequence ($p > 0.1$), period ($p > 0.05$), or treatment ($p > 0.05$) effects.

3. Individual subject ratios (Trt. A/Trt. B) for pharmacokinetic parameters are shown in Table 5.

D. Analytical

VIII. Comments

A. Product Information

1. On p. 95 and 429 the test product lot# is stated as "SB026". On p. 169, 193-4, 438, and 440-1, the test product lot# is stated as "SB0026".

2. The sponsor did not provide CV's for the dissolution data or the analytical method.

B. Clinical

1. The sponsor did not provide case report forms, adverse reaction reports, or other clinical raw data from the study.

C. Pharmacokinetics/Statistics

1. **KEL values:** Table 6 is a summary of literature data regarding the half-life of 5MTHF and the decline of 5MTHF in plasma or serum. In the majority of cases, the semilog plots appear to indicate that 5MTHF is eliminated in a monoexponential manner. However, in two cases, the decline may be biexponential. Due to the small number of sampling times in most of these studies, conclusions are not firm.

Two KEL values (S14 and 31, Trt. A) are unacceptable since the terminal data point "bounces up".

For the remaining 62 curves, the reviewer's conclusions from visual inspection of the semilog plots provided by the sponsor are as follows:

- 18 of the curves appear to exhibit monophasic serum level decline of 5MTHF
- 44 of the curves appear to have some degree of biphasic serum level decline for 5-MTHF with slope changes occurring at 12-16 hours
- In 33 cases, the correlation coefficients (r) for the KEL values were < 0.95 . In 3 cases, $r < 0.90$ (excluding the two values deemed unacceptable above). For all these cases, the curves exhibited biphasic serum level decline.
- The sponsor used start times for KEL determination ranging from 6-12 hr.

D. Analytical

IX. Waiver Request

1. The sponsor requests waiver of in vivo bioequivalence study requirements for its lower strength test product leucovorin calcium 5 mg tablets. This request is based on formula proportionality to the 25 mg strength, acceptable dissolution testing results, and an acceptable biostudy for the higher strength test product.
2. From Table 1 it is evident that both strengths contain the same quantities of excipients except for the filler ingredient.

X. Deficiencies

1. On p. 95 and 429 the test product lot# is stated as "SB026". On p. 169, 193-4, 438, and 440-1, the test product lot# is stated as "SB0026". The sponsor should state the correct lot number for the test biostudy lot.
2. The Division of Bioequivalence guidance for leucovorin calcium tablets requests that the sponsor determine the ratio of D:L isomers in both the test and reference biostudy lots, and that these ratios should not differ by more than 5%. This information was not submitted.
3. For the dissolution data, the sponsor did not provide CV's for the mean dissolution values at each sampling time or the analytical method.
4. The sponsor must provide case report forms, adverse reaction reports, and all other relevant clinical raw data from the study.

5. The majority of the semilog plots provided by the sponsor appear visually to indicate that the serum level decline of 5MTHF is biphasic with slope changes occurring around 12-16 hours postdose. It is noted that the sponsor used start times for KEL determination ranging from 6-12 hours. It is also noted that in 33 cases, the correlation coefficient is < 0.95 . The sponsor should comment on the suitability of using data points earlier than the 12-16 hr range as part of the terminal elimination phase.

XI. Recommendations

1. The bioequivalence study conducted by Par Pharmaceutical on its leucovorin calcium 25 mg tablet, lot #SB0026, comparing it to Burroughs Wellcome's Wellcovorin® 25 mg tablet, lot #2T2271, has been found incomplete by the Division of Bioequivalence due to deficiencies 1-8 stated above.

2. The sponsor is requested to provide a 3.5" diskette containing the pharmacokinetic data from the study. Files should be configured as follows:

File #1: subj seq per trt auc aucinf cmax tmax kel $t_{1/2}$

File #2: subj seq per trt conc1-conc14

File #3: subj seq per trt conc1-conc14

File #2 represents the actual data, and File #3 is the baseline-adjusted data.

3. The sponsor should resubmit its request for waiver of in vivo bioequivalence study requirements for its lower strength test

product leucovorin calcium 5 mg tablets with the responses to the deficiency comments above.

4. The firm should be informed of deficiencies 1-8 and recommendations 1-3.

James D. Henderson, Ph.D.
Review Branch II
Division of Bioequivalence

RD INITIALED RPATNAIK
FT INITIALED RPATNAIK _____

Concur: _____ Date 3/31/95
Rabindra N. Patnaik, Ph.D.
Acting Director
Division of Bioequivalence

JDH/crc/3-31-95/74544

cc: ANDA #74-544 (original, duplicate), HFD-600 (Hare), HFD-630, HFC-130 (JAllen), HFD-344 (CViswanathan), HFD-655 (Patnaik, Henderson), Drug File, Division File

Table 1 - Formulations of the Test Products

Ingredient	25 mg - mg/tablet	5 mg - mg/tablet
leucovorin calcium USP ¹	27.0	5.4
anhydrous lactose NF		
microcrystalline cellulose NF		
croscarmellose sodium NF		
colloidal silicon dioxide NF		
magnesium stearate NF		

¹ According to the labeling for Wellcovorin® (PDR, 1994, p. 721), 5.4 and 27.0 mg of leucovorin calcium contain 5 or 25 mg of leucovorin, respectively.

Table 2. In Vitro Dissolution Testing

Drug (Generic Name): leucovorin calcium
Dose Strength: 5 & 25 mg
ANDA No.: 74-544
Firm: Par
Submission Date: 9/16/94
File Name: 74544SDW.994

I. Dissolution Testing (USP Method):

USP XXII Basket: Paddle: X RPM: 50
No. Units Tested: 12
Medium: water Volume: 900 mL
Specifications: NLT 30 min
Reference Drug: Wellcovorin® (Burroughs Wellcome)
Assay Methodology: not stated

II. Results of In Vitro Dissolution Testing:

Sampling Times (Minutes)	Test Product Lot #SB0027 Strength (mg) 5			Reference Product Lot #1W1638 exp 11/94 Strength (mg) 5		
	Mean %	Range	%CV	Mean %	Range	%CV
15	88.5		-	94.5		-
30	96.2		-	99.3		-
45	97.0		-	97.0		-
60	97.6		-	97.3		-
Sampling Times (Minutes)	Test Product Lot #SB0026 Strength (mg) 25			Reference Product Lot #2T2271 Strength (mg) 25 exp 8/95		
	Mean %	Range	%CV	Mean %	Range	%CV
15	95.9		-	97.5		-
30	98.7		-	99.3		-
45	99.5		-	99.6		-
60	100.0		-	99.2		-

**Table 3 - Mean Reported Serum Concentrations of 5MTHF
(Baseline-Adjusted, N = 32, ng/mL)**

<u>Time</u> (hr)	<u>Trt. A</u> (mean)	(test) CV%	<u>Trt. B</u> (mean)	(ref.) CV%	<u>% diff.</u>
-1.0	0.00	-	0.00	-	-
0.5	67.17	48	74.81	48	-10.21
1	167.17	38	183.91	28	-9.10
1.5	221.07	33	225.19	22	-1.83
2	235.35	34	237.55	23	-0.93
2.5	237.03	34	230.63	22	2.78
3	220.49	36	218.13	24	1.08
4	179.47	42	181.31	34	-1.01
6	72.41	38	73.77	38	-1.84
8	46.48	28	48.37	30	-3.91
10	34.31	27	34.73	27	-1.21
12	25.68	25	25.86	27	-0.70
16	17.47	27	17.47	27	0.00
24	12.94	29	12.69	30	1.97

Trt. A = leucovorin calcium (Par), 1 X 25 mg tablet

Trt. B = Wellcovorin® (Burroughs Wellcome), 1 X 25 mg tablet

Table 4 - Mean Reported Pharmacokinetic Parameters of 5MTHF
(Baseline-Adjusted Means¹, N = 32)

Parameter	Trt. A (mean)	CV (%)	Trt. B (mean)	CV (%)	% diff. or Ratio ²	90% CI
AUC ₀₋₂₄ ³	1438.4	28	1458.5	21	-1.38	90.4-106.8
logAUC ₀₋₂₄	-	-	-	-	0.968	88.3-106.2
AUC _{INF}	1594.3	28	1630.6	22	-2.23	89.7-105.8
logAUC _{INF}	-	-	-	-	0.963	88.1-105.3
C _{MAX} (ng/mL)	252.77	35	253.48	23	-0.28	90.1-109.3
logC _{MAX}	-	-	-	-	0.963	87.1-106.5
T _{MAX} (hr)	2.31	24	2.20	27	5.00	-
K _{EL} (hr ⁻¹)	0.0857	15	0.0847	25	1.18	-
t _{1/2} (hr)	8.26	15	8.97	40	-7.92	-

¹ For this balanced study, arithmetic means and least-squares means are identical.

² For untransformed parameters, the % difference of arithmetic means = $(A - B) * 100 / B$. For log-transformed parameters, the ratio of LS Geometric means = antilog(estimate) from the ANOVA.

³ units for AUC: ng*hr/mL

Trt. A = leucovorin calcium (Par), 1 X 25 mg tablet

Trt. B = Wellcovorin® (Burroughs Wellcome), 1 X 25 mg tablet

Table 5 - Individual Ratios (A/B) for Pharmacokinetic Parameters

<u>Subject</u>	<u>AUCD-24</u>	<u>AUCINF</u>	<u>C_{MAX}</u>
1			
2			
3			
4			
5			
6			
7			
8			
9			
10			
11			
12			
13			
14			
15			
16			
17			
18			
19			
20			
21			
22			
23			
24			
25			
26			
27			
28			
29			
30			
31			
32			
< 75%	4	5	8
75-125%	21	19	16
> 125%	7	8	8

Table 6 - Literature Data for Half-life of 5-MTHF

Ref	Dose	Subjects	Assay	$t_{1/2}$ (hr)	Comment
1	rac-LV ¹ iv 25 mg, N=6 50 mg, N=12 100 mg, N=6 rac-LV po 25 mg, N=6 50 mg, N=11 100 mg, N=10	normal	HPLC	3.9 3.7 3.8 5.2 3.2 3.0	2
2	rac-LV, 50 mg iv	normal	HPLC	3.9	2
3	rac-LV, 25 mg iv, N=36 im, N=34 po, N=34	normal	micro	6.5 6.4 6.0	-
4	rac-LV, iv 100 mg, N=28 250 mg, N=28	normal	HPLC	5.1 5.0	3 2
5	rac-LV 25 mg po, N=8	normal	micro	-	4,5
6	folic acid, N=10 25 mg, iv & po 125 mg, iv & po	normal	radio- enzymatic	- - -	2
7	rac-LV, iv ⁶ , N=7	patient	HPLC	7	2,5
8	rac-LV 25 mg po, N=36	normal	micro	5.2, 5.3	5
9	rac-LV 25 mg po, N=35 30 mg iv, N=33 30 mg po, N=33	normal	RIA	6.9 6.9 5.8	-

- 1 rac-LV = racemic leucovorin calcium
 - 2 plasma level decline from semilog plot appears
 - 3 monoexponential
 - 4 for dose = 100 mg, plot appears biexponential, but only five
 - 5 data points are present
 - 6 apparent biexponential serum level decline, sampling to 24
- hr
- (S)-(-)-5-MTHF
- loading dose 200 mg/m² followed by 400 mg/m² 2-hr infusion

REFERENCES

1. Straw JA, Szapary D, Wynn WT. Pharmacokinetics of the diastereomers of leucovorin after intravenous and oral administration to normal subjects. *Cancer Res* 1984;44:3114-9.
2. Payet B, Fabre G, Tubiana N, Cano J. Plasma kinetic study of folinic acid and 5-methyltetrahydrofolate in healthy volunteers and cancer patients by high-performance liquid chromatography. *Cancer Chemother Pharmacol* 1987;19:319-25.
3. McGuire BW, Sia LL, Leese PT, Gutierrez ML, Stokstad EL. Pharmacokinetics of leucovorin calcium after intravenous, intramuscular, and oral administration. *Clin Pharm* 1988;7:52-8.
4. Thyss A, Milano G, Etienne MC, Paquis P, Roche JL, Grellet P, Schneider M. Evidence for CSF accumulation of 5-methyltetrahydrofolate during repeated courses of methotrexate plus folinic acid rescue. *Br J Cancer* 1989;59:627-30.
5. Zittoun J, Tonelli AP, Marquet J, de Gialluly E, Hancock C, Yacobi A, Johnson JB. Pharmacokinetic comparison of leucovorin and levoleucovorin. *Eur J Clin Pharmacol* 1993;44:569-73.
6. Schmitz JC, Stuart RK, Priest DG. Disposition of folic acid and its metabolites: a comparison with leucovorin. *Clin Pharmacol Ther* 1994;55:501-8.
7. Mader RM, Steger GG, Rizovski B, et al. Pharmacokinetics of rac-leucovorin vs [S]-leucovorin in patients with advanced gastrointestinal cancer. *Br J Clin Pharmacol* 1994;37:243-8.
8. De Vito JM, McGuire BW, de Lap RJ, Weiss AI. Bioequivalence of two leucovorin calcium tablet formulations. *Drug Intell Clin Pharm* 1989;23:153-4.
9. De Vito JM, Kozloski GD, Tonelli AP, Johnson JB. Bioequivalence of oral and injectable levoleucovorin and leucovorin. *Clin Pharm* 1993;12:293-9.